

WHAT IS CLAIMED IS:

1. A method for producing mutant polynucleotides comprising:
producing polynucleotides by blocking or interrupting a polynucleotide
synthesis or amplification process with a member selected from the group consisting of
5 UV light, one or more DNA adducts, DNA intercalating agents, DNA binding proteins,
triple helix forming agents, competing transcription polymerase, chain terminators, and
polymerase inhibitors or poisons, said member being capable of blocking or interrupting
synthesis or amplification of a polynucleotide to provide a plurality of polynucleotides
due to said polynucleotides being in various stages of synthesis or amplification, and
10 subjecting said polynucleotides to an amplification procedure to amplify one
or more of the polynucleotide or polynucleotides.
2. A process for producing mutant polynucleotides by a series of steps
comprising:
(a) producing oligonucleotides by blocking or interrupting a polynucleotide
15 synthesis or amplification process with at least one member selected from the group
consisting of UV light, one or more DNA adducts, DNA intercalating agents, chain
terminators, and/or polymerase inhibitors or poisons, wherein said member is capable of
blocking or interrupting polynucleotide synthesis or amplification and provide a plurality
of polynucleotides due to their being in various stages of synthesis or amplification.
20 (b) denaturing the resulting single or double stranded oligonucleotides to
produce a mixture of single-stranded polynucleotides, optionally separating the
polynucleotides into pools of polynucleotides having various lengths, and further
optionally subjecting said polynucleotides to a priming and amplification procedure to
amplify one or more oligonucleotides comprised by at least one of the polynucleotide
25 pools;
(c) incubating a plurality of said polynucleotides or at least one pool of
said polynucleotides with a polymerase under conditions which result in annealing of

Acet⁺ said single-stranded polynucleotides at regions of identity between the single-stranded polynucleotides and formation of mutagenized double stranded polynucleotide chain;

(d) repeating steps (c) and (d);

(e) expressing at least one mutant polypeptide from said polynucleotide chain,
5 or chains; and

(f) screening said at least one mutant polypeptide for a useful activity.

3. A process according to claim 2, wherein said adduct is member selected from the group consisting of: UV light; (+)-CC-1065; (+)-CC-1065-(N3-Adenine); a N-acetylated or deacetylated 4'-fluoro-4-aminobiphenyl adduct capable of inhibiting DNA
10 synthesis, or a N-acetylated or deacetylated 4-aminobiphenyl adduct capable of inhibiting DNA synthesis; trivalent chromium; a trivalent chromium salt; a polycyclic aromatic hydrocarbon ("PAH") DNA adduct capable of inhibiting DNA replication; 7-bromomethyl-benz[a]anthracene ("BMA"); tris(2,3-dibromopropyl)phosphate ("Tris-BP"); 1,2-dibromo-3-chloropropane ("DBCP"); 2-bromoacrolein (2BA); benzo[a]pyrene-
15 7,8-dihydrodiol-9-10-epoxide ("BPDE"); a platinum(II) halogen salt; N-hydroxy-2-amino-3-methylimidazo[4,5-f]-quinoline; N-hydroxy-2-amino-1-methyl-6-phenylimidazo[4,5-f]-pyridine, DNA intercalating agents, DNA binding proteins, triple helix forming agents, competing transcription polymerases, chain terminators, and polymerase inhibitors or poisons.

20 4. A process according to claim 2, wherein said DNA adduct is a member selected from the group consisting of UV light, (+)-CC-1065 and (+)-CC-1065-(N3-Adenine).

5. A process according to claim 4, further comprising heating said polynucleotides and removing the DNA adduct, or adducts from said polynucleotide or polynucleo-
25 tide pools.

add.

6. A method for expressing a polypeptide comprising producing a polynucleotide according to claim 2 and comprising the further steps of cloning said polynucleotide into a vector or an expression vehicle and expressing said polypeptide.

7. A vector or an expression vehicle including a polynucleotide produced according to claim 2.

8. A polypeptide comprising at least one sequence segment expressed from a polynucleotide produced by the method according to claim 2.

Add A3

add C2

0044949260